

ThermoFisher
S C I E N T I F I C

Matlab + Compressive Sensing + Scanning Electron Microscopy

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The world leader in serving science

Overview – How we use Matlab for Big Data Acquisition

- **Life Science, Electron Microscopy**
- **Matlab Integration: Image Acquisition Stability**
- **Compressive Sensing Basics**
- **Image Acquisition with CS**
- **Matlab: Reconstruction Algorithms**
- **Matlab Integration: Integration to Acquire Big Data**



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THE GRIFFITH OLUB MICROSCOPE.



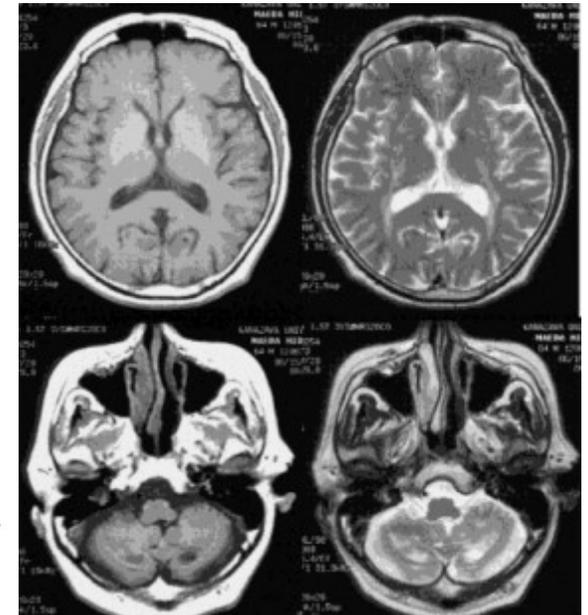
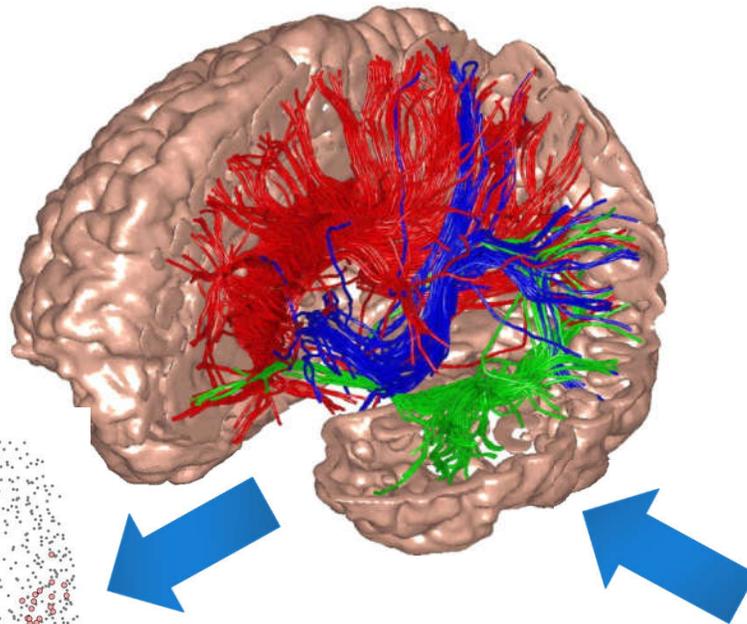
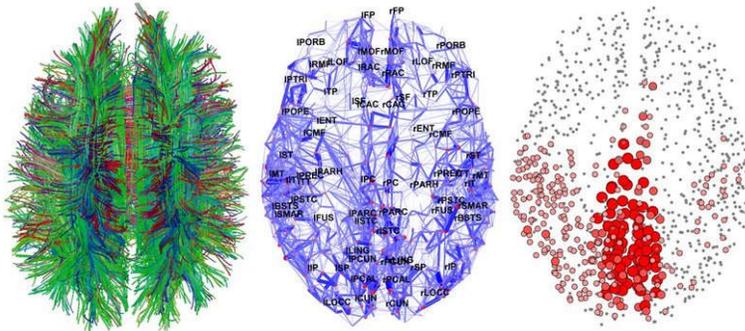
- Increased interest to understand the brain functionality

From Connections to Cognition

Human Connectome Project
(USA launched 2010 to map
wiring human brain)

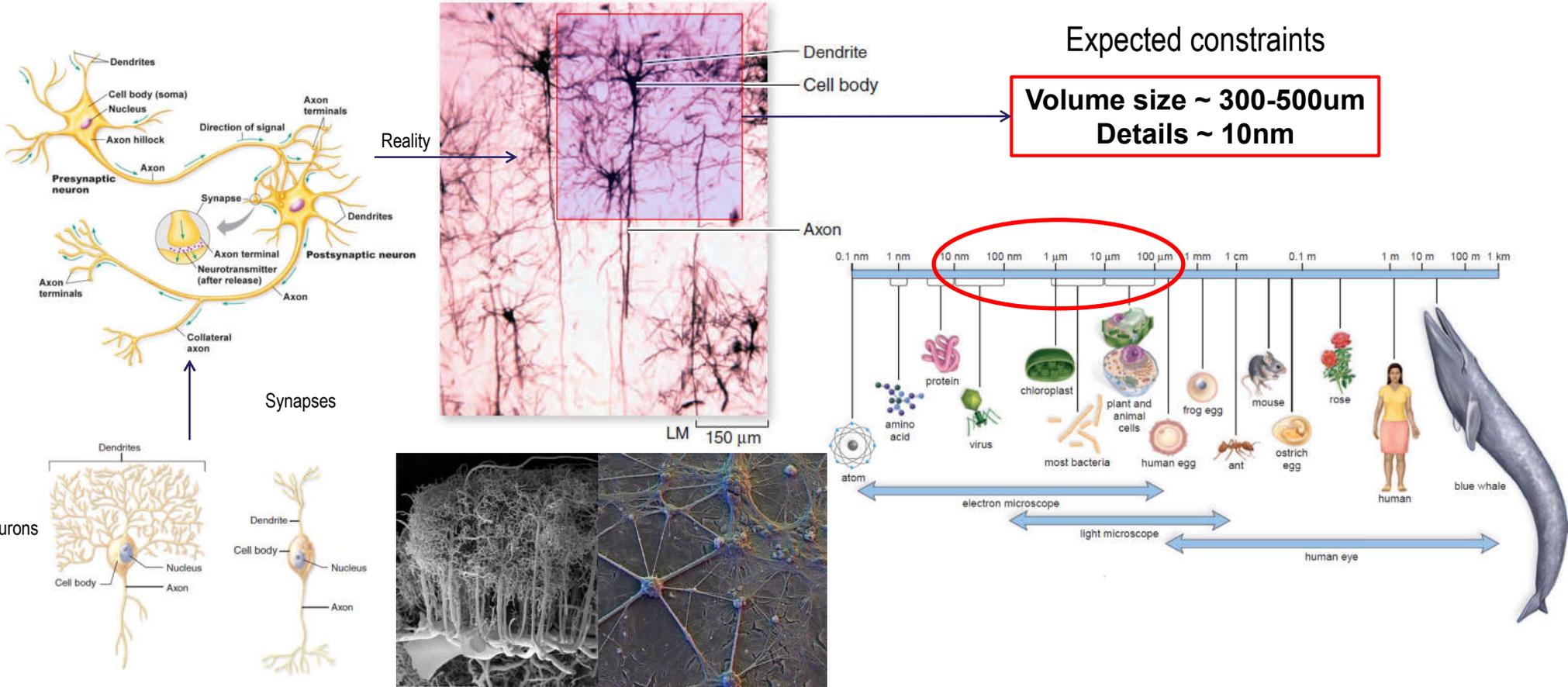
Connectome

Sum of all brain's connections



Brain investigation example

Neurons and their connections - synapses



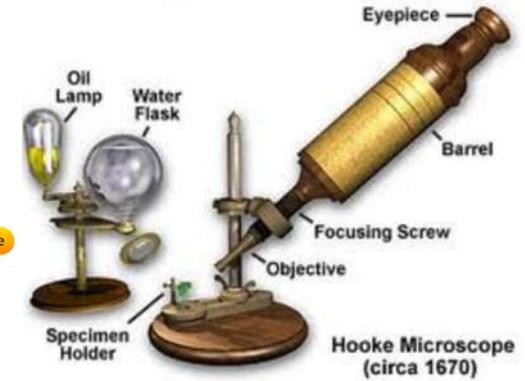
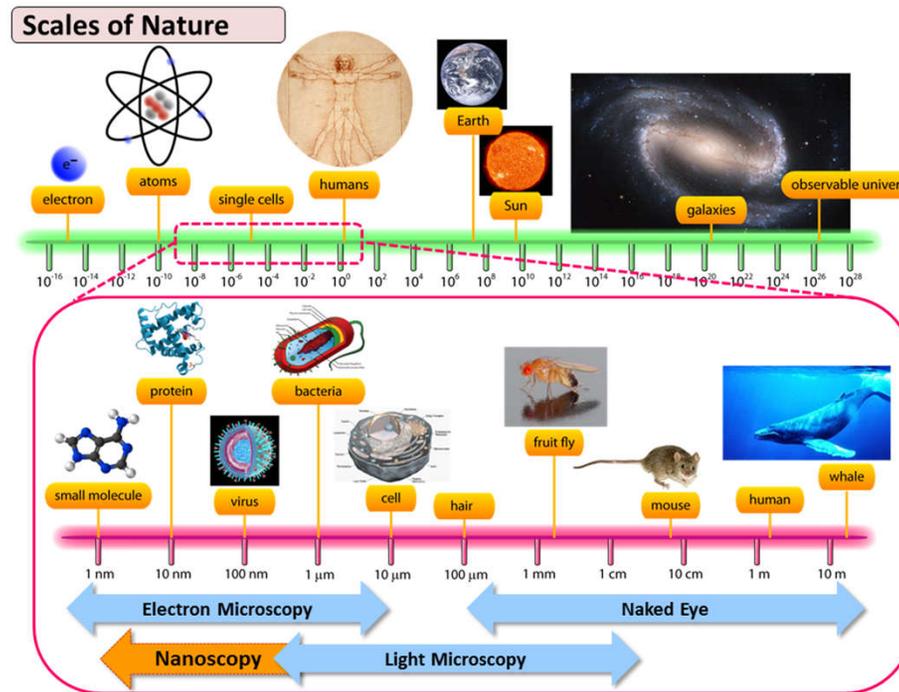
Which tool for Imaging?



SEM 1937 – von Ardenne



TEM 1931 – Knoll, Ruska



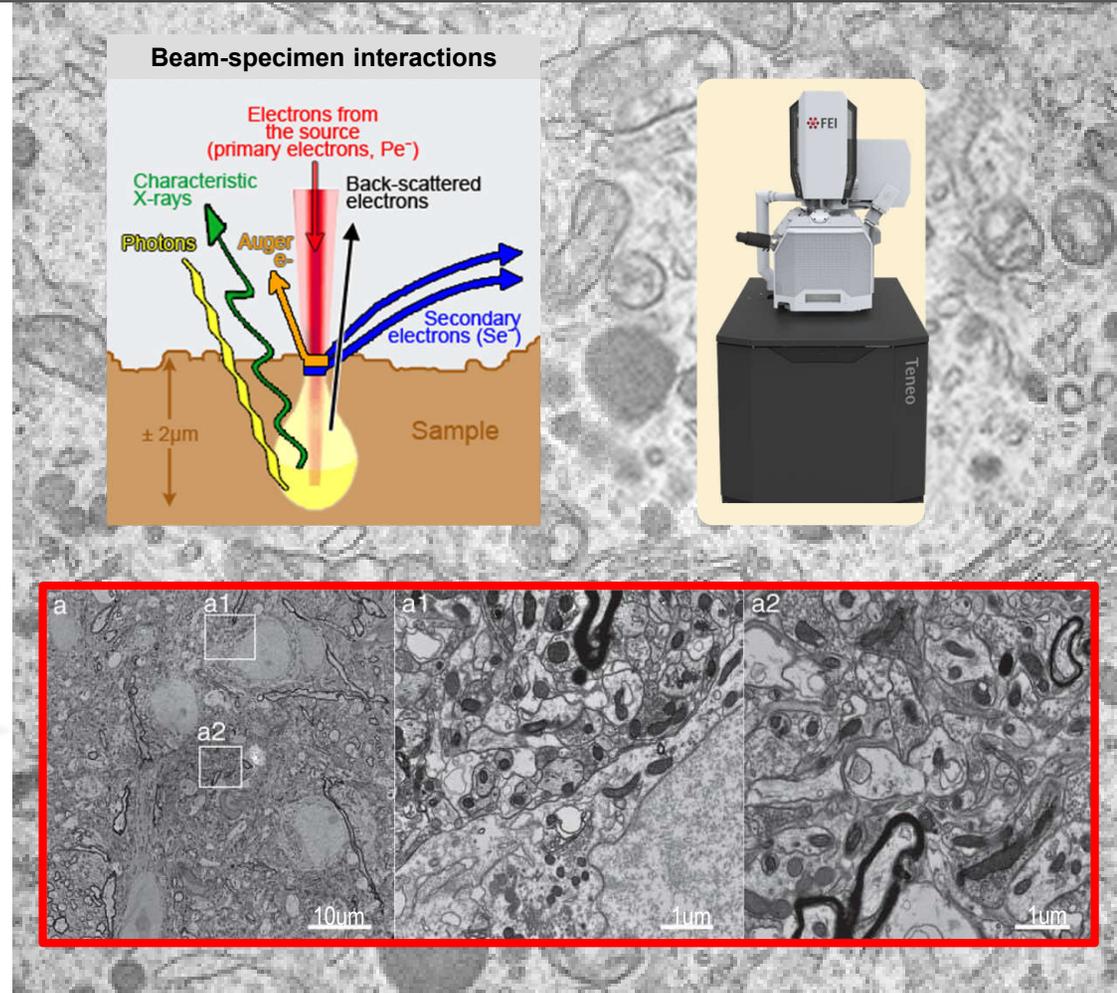
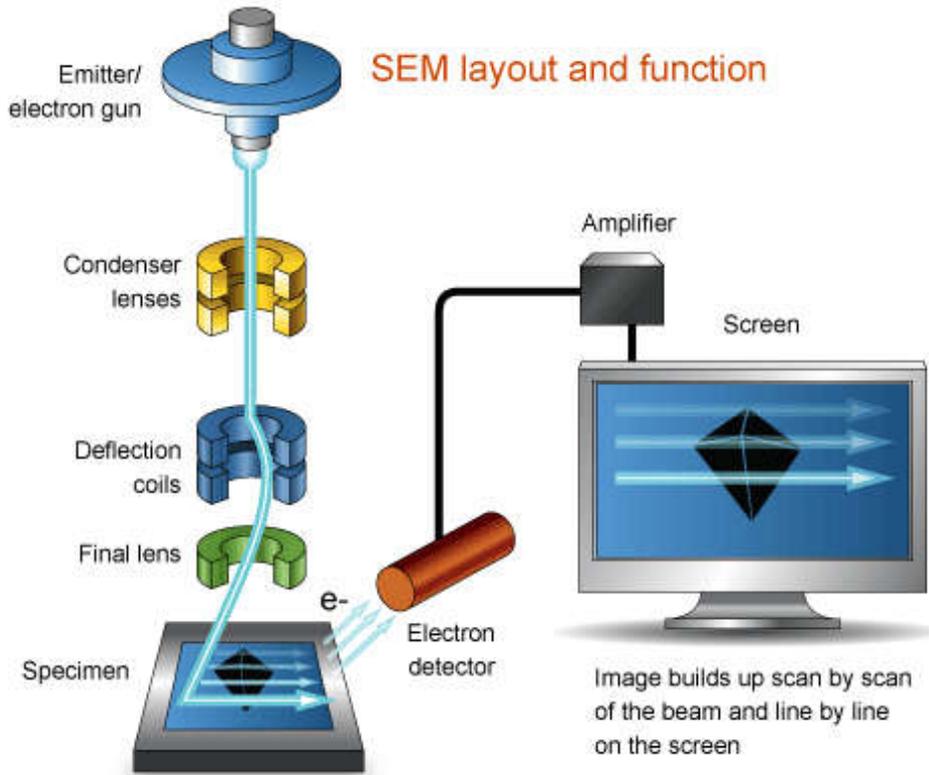
Hooke Microscope (circa 1670)

This state-of-the-art microscope contains accessories for DIC, fluorescence, polarized light, phase contrast, and photomicrography using several film formats and digital image capture.



The Olympus Provis AX-70 (circa 1998)

Very Short introduction to Scanning Electron Microscopy



Brain Reverse Engineering

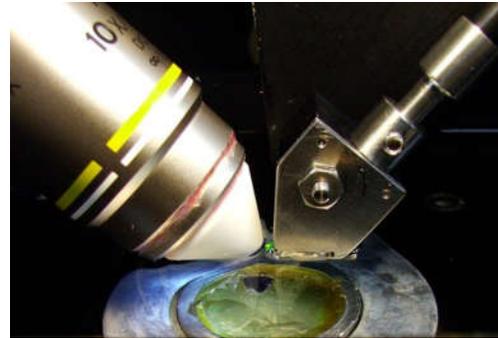
- Reverse engineering is one of the most important techniques



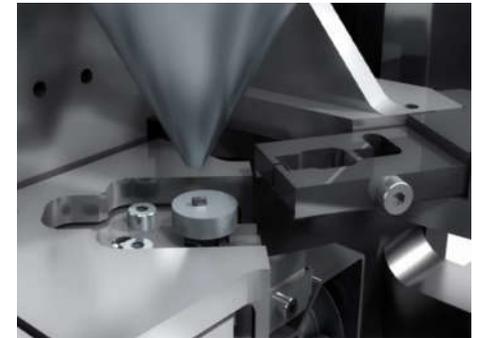
Knife
1mm slice



Blade
70um slice



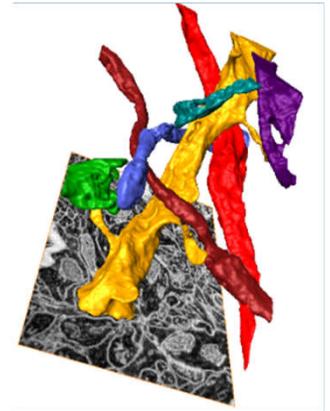
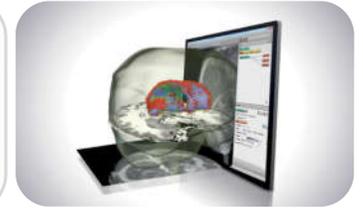
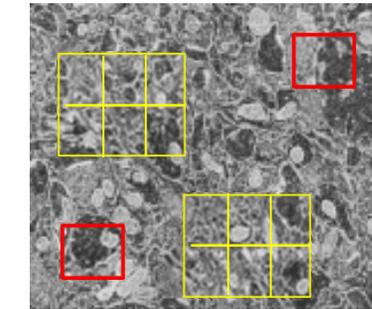
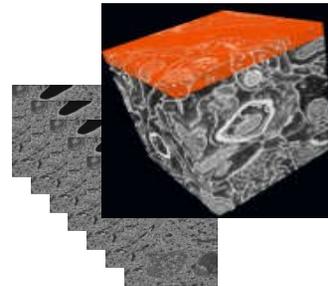
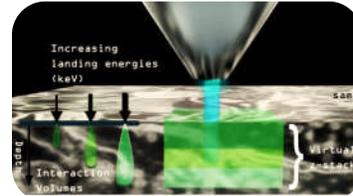
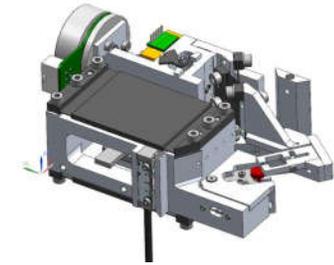
Knife-edge, SEM
300-500nm slice
full mouse brain about 100 hours



Diamond knife, SEM
25-50nm slice

Data acquisition workflow

VolumeScope



Teneo SEM

Microtome

**Multi-Energy
Reconstruction**

MAPS

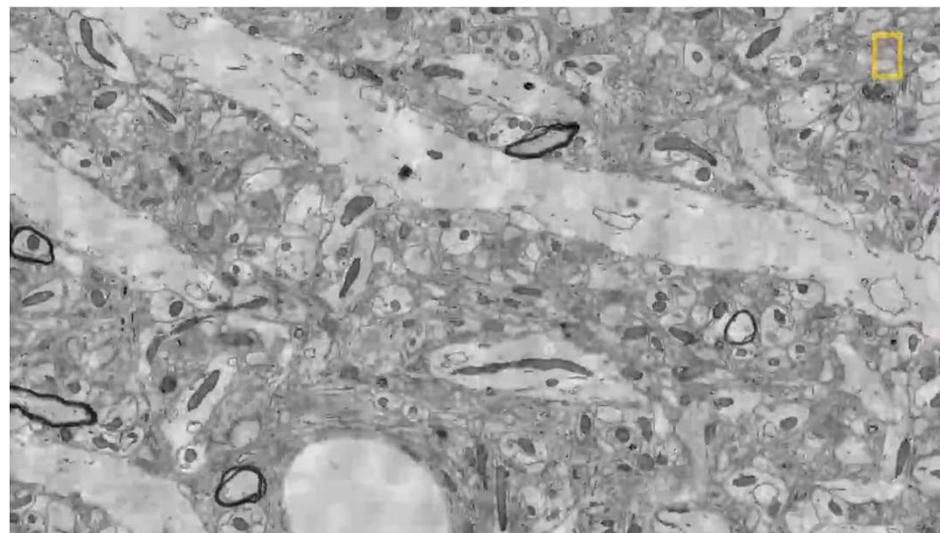
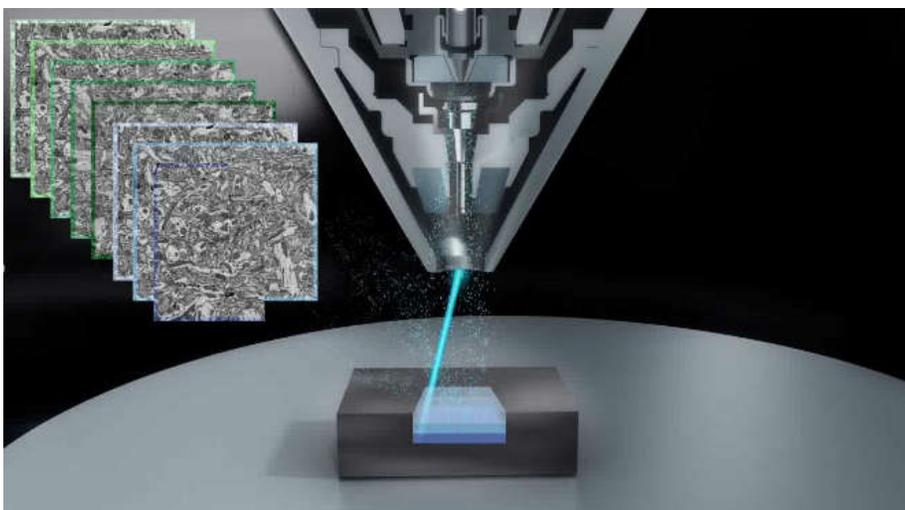
AMIRA

Hardware

Software

Data acquisition workflow overview - movies

VolumeScope data acquisition



Neuron reconstruction

(from National Geographic, Lichtman)

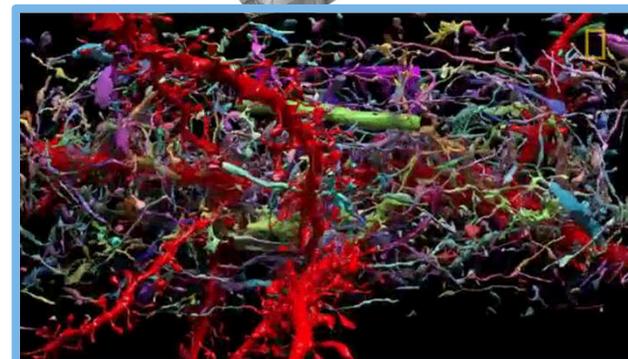
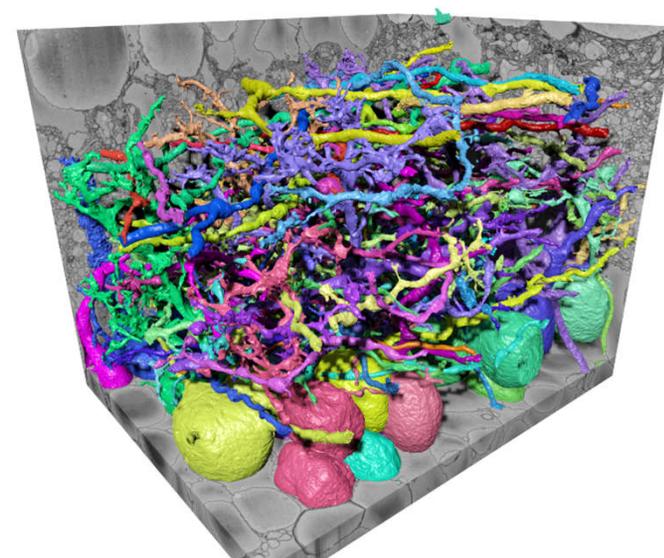
Acquisition timing example

**Volume size ~ 300-500um
Details ~ 10nm**

Acquisition	Dwell time	100ns	1us
Single section image	Pixel resolution	32-64k	32-64k
	Data size	2 – 8 GB	2 – 8 GB
	Acquisition time	2-8 m	20-80 m
Full 3D volume	Data size	64 - 512 TB	64 - 512 TB
	Acquisition time	38 – 304 d	380 – 3040 d

- No cutting
- No tiling involved – No stage movements
- No auto-functions - No processing
- Single 16bits data instance
- No failure

MPI:
Denk, Helmstaedter:
13kx50kx50k = 33TP
8 weeks acquisition;
2years to reconstruct



Overview – How we use Matlab for Big Data Acquisition

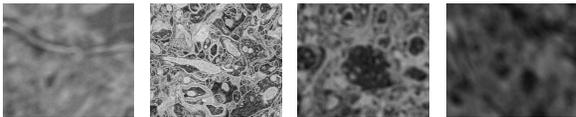
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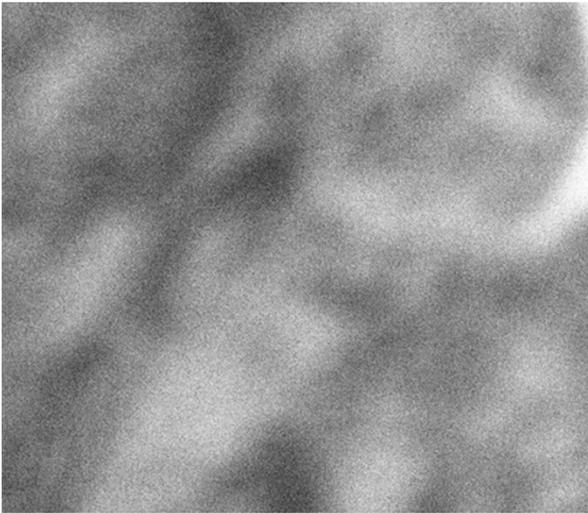
Why do we need auto-functions?

Different imaging conditions (beam energy, working distance, ...)

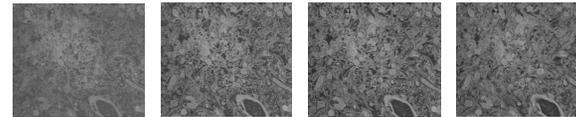
Without any adaptation



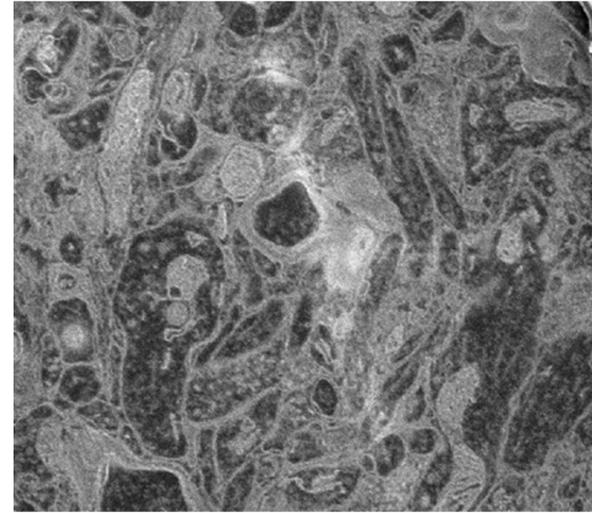
Reference



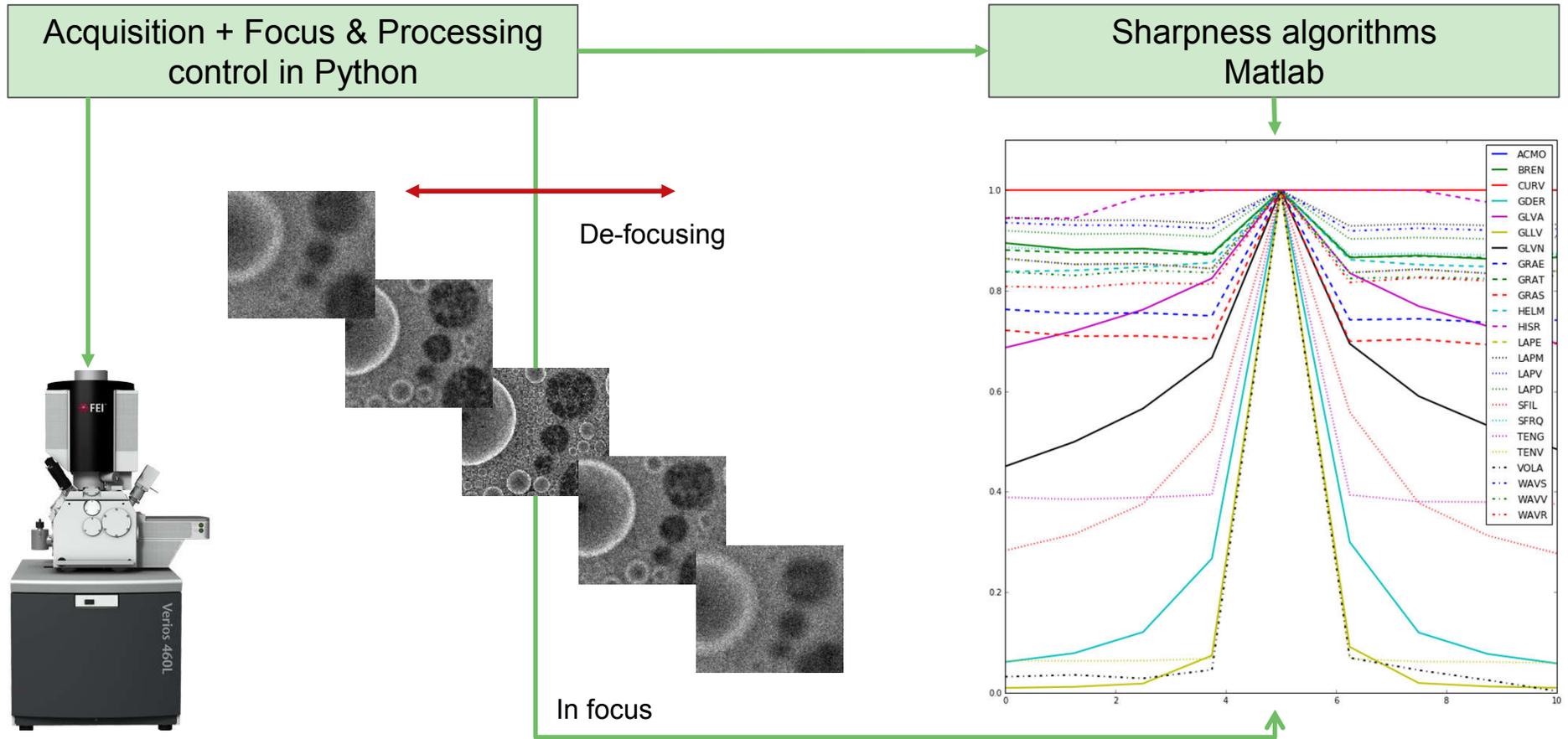
Compensation after auto-functions



Reference



Matlab to test different focusing algorithms



Matlab inline in Python notebook with other languages

Code

```
In [1]: # XT MPC connection
import pycsharpbridge as pycsharp

ip = get_ipython()
if connect_to_xt:
    ip = get_ipython()
    pycsharp.load_ipython_extension(ip, csharp=kernelExe, socket_addr=kernelSocket)
```

C#

```
Out[1]: Test socket is_kernel_running :tcp://mpc-d0000:9002
Req:{"cmd": "connect"}
.Connected to already running csharp kernel
```

```
In [2]: # load matlab magic extension
import pymatbridge as pymat

ip = get_ipython()
pymat.load_ipython_extension(ip)
# get the matlab magic module
magic = ip.magics_manager.registry['MatlabMagics']
```

Matlab

```
Out[2]: Starting MATLAB on ZMQ socket tcp://Matlab-station:54400
Send 'exit' command to kill the server
.....MATLAB started and connected!
```

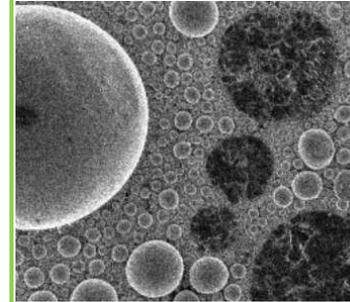
```
In [3]: # Image acquisition (C#) and sharpness calculation (matlab)
```

Matlab + C#

```
wd = 3.5e-3
%csharp MakeWDSnapshot(wd, 0); -i wd
%csharp ArtImage image = GetImage(0); -o image
display(image.resize((256,221)))

# get sharpness from matlab
%matlab ; -i image
magic.Matlab.run_code(matlabcode)
%matlab ; -o compSharpness
print(NiceFormat(compSharpness))
```

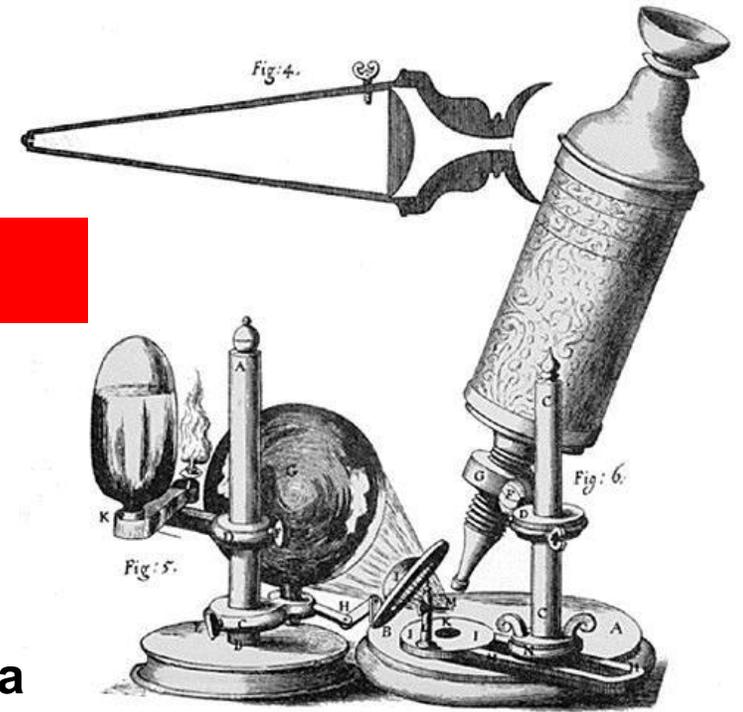
Output



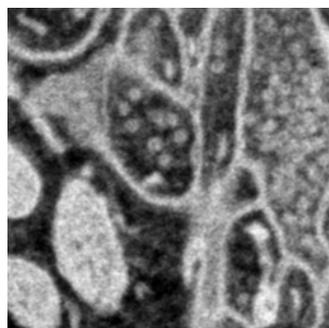
ACMO:0.06947	64770
BREN:0.0057909	4315.3128
CURV:0.068051	26
GDER:0.037985	3.211550742768305e+35
GLVA:0.021402	37.0071
GLLV:0.031385	7875.9058
GLVN:0.0023444	7.5538
GRAE:0.0051892	2851.0375
GRAT:0.0055283	41.2238
GRAS:0.0024659	1261.0714
HELM:0.034528	1.1402
HISE:0.0067125	0
HISR:0.0010029	219
LAPE:0.0085796	8689.4197
LAPM:0.0062981	101.0271
LAPV:0.0047512	8689.4581
LAPD:0.011362	176.2448
SFIL:0.046259	4.483135721068581e+17
SFRQ:0.0067394	46.3789
TENG:0.006919	19174.2075
TENV:0.007676	358916026.9115
VOLA:0.0054004	5.6804
WAVS:0.23817	30.7009
WAVV:0.045032	97.8731
WAVR:0.099396	0.95287

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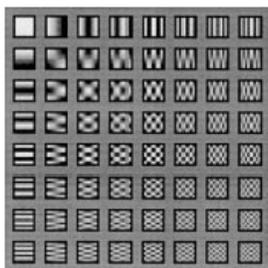
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Compressible vs Sparse Signals



Original



DCT basis

Compressed images – Compressible images – Sparse images



Frac: 0.400



Frac: 0.150

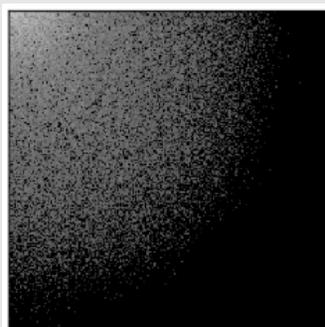


Frac: 0.050

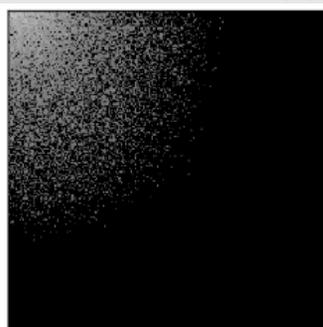


Frac: 0.010

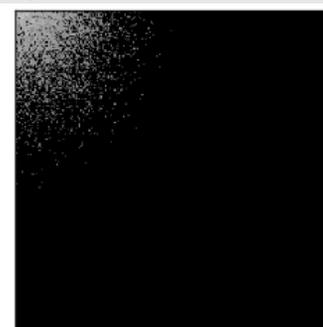
Cropped spectrums



Frac: 0.400



Frac: 0.150



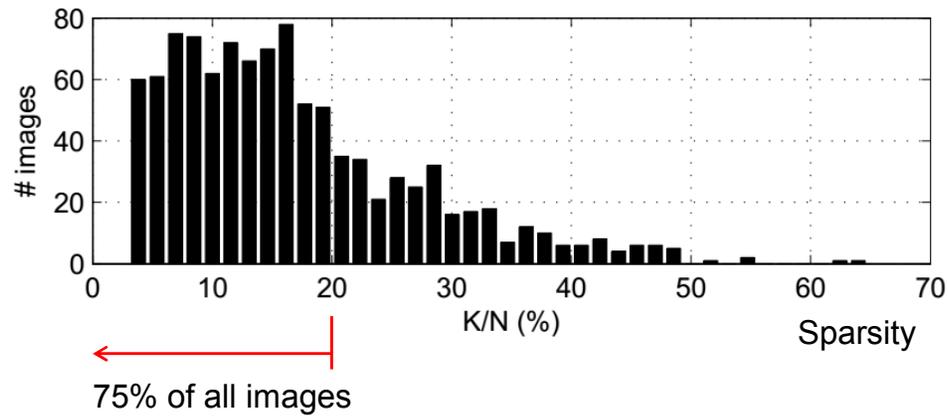
Frac: 0.050



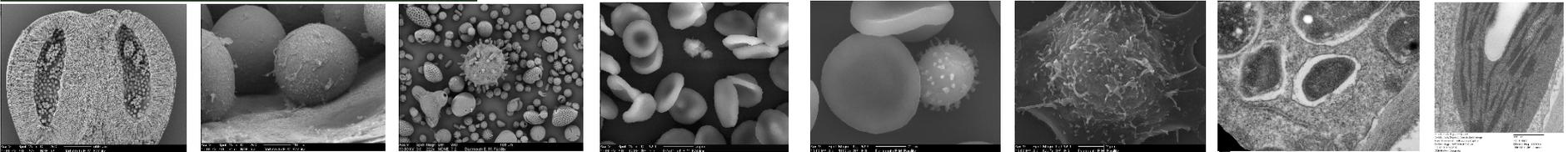
Frac: 0.010

Compressive sensing – motivation from JPEG

- How sparse is the microscopy image?

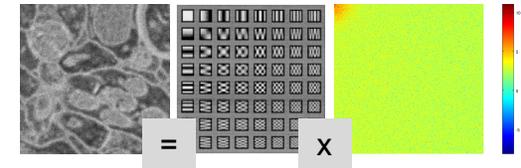
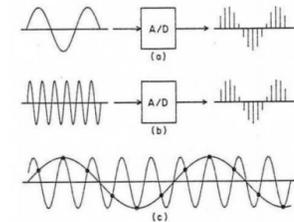


Dartmouth College Electron Microscope Facility



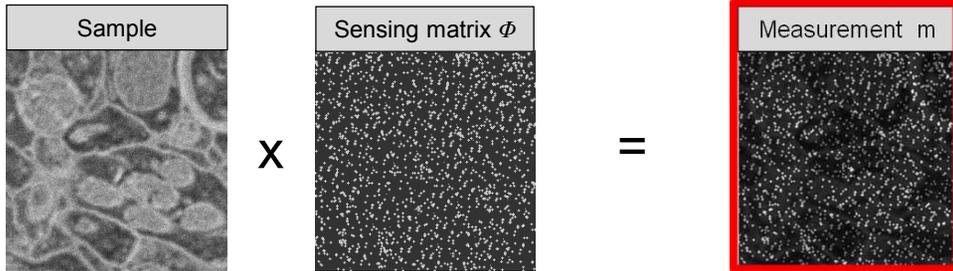
Signal reconstruction for Compressive sensing

- Signal reconstruction from sampling measurements
- Nyquist-Shannon theorem
samples depends on the signal's frequencies
- Candes-Tao-Donoho (2004-2006)
samples depends on the signal's sparsity

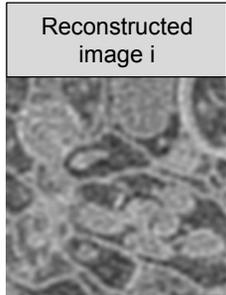
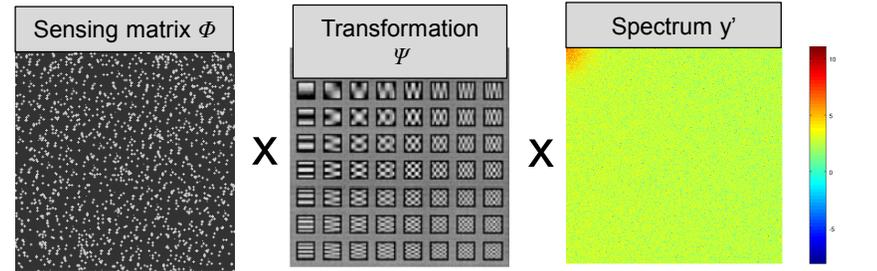


Compressive sensing principle

Measurement process



Reconstruction process



$$i = \Psi y$$

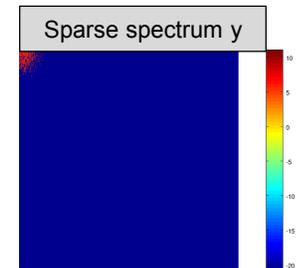
$$\min \|y\|_0$$

such that

$$m = \Phi \Psi y$$



Iterative searching for...



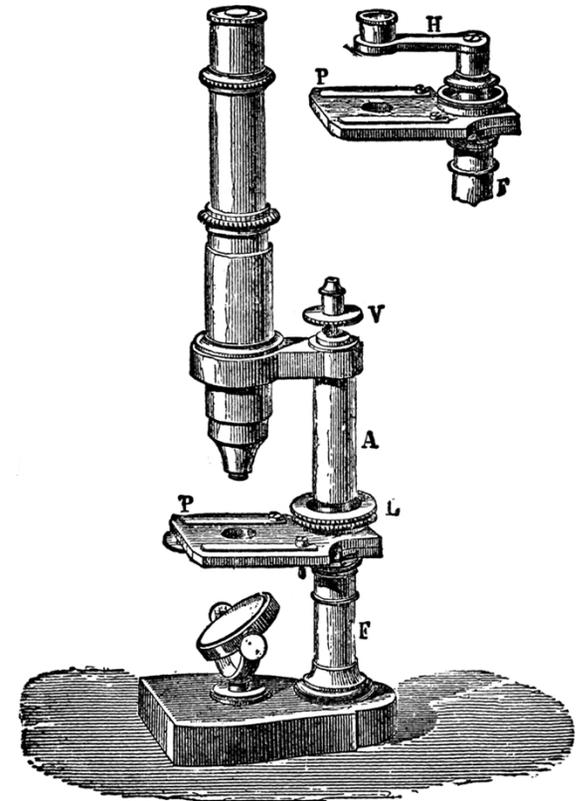
Sparse undetermined system
 ill-posed problem
 (uniqueness is lacking)

Sparse recovery:

- Non-linear optimization that promotes sparsity
- Find the sparsest solution that is consistent with measured data

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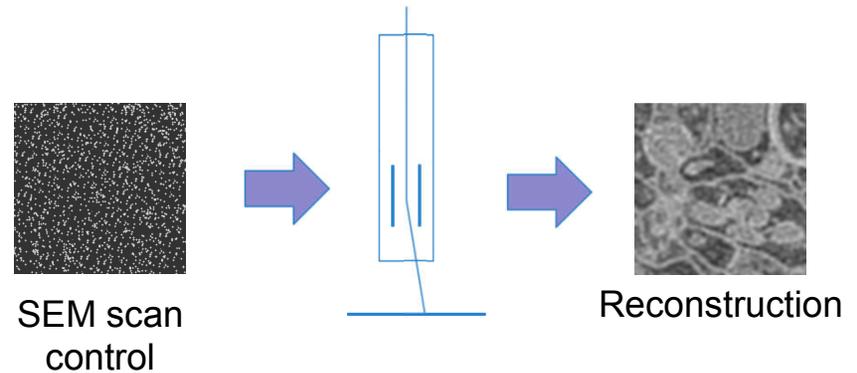
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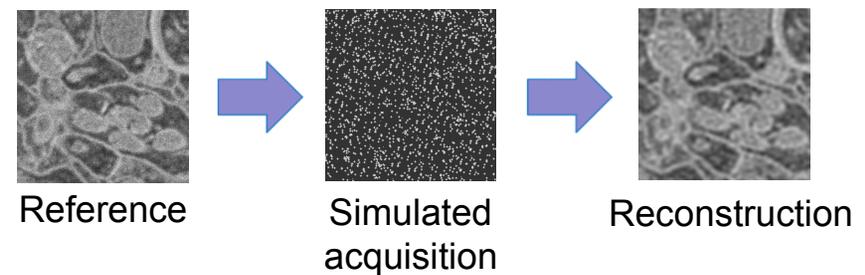
What we need for the Compressive Sensing for EM?

Promise of high speed and low dose imaging.

- Sparse scanning on SEM/STEM



- Reconstruction algorithms

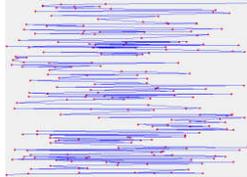


Scanning strategies

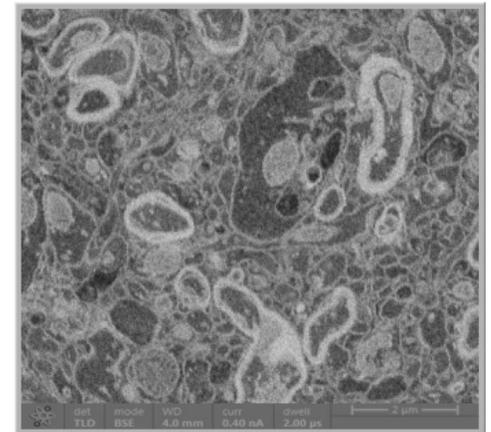
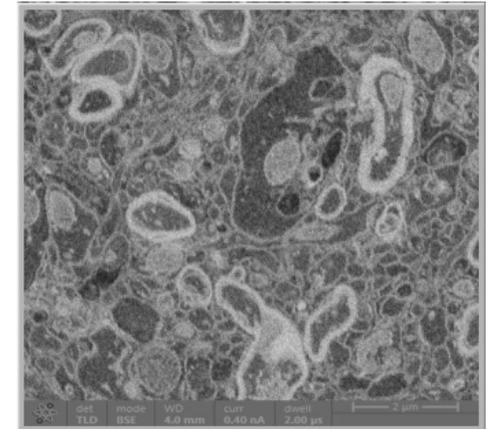
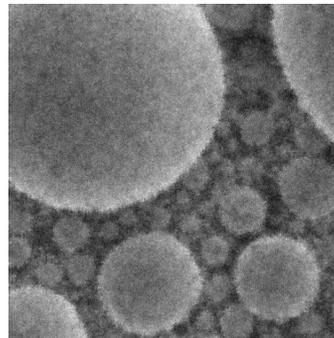
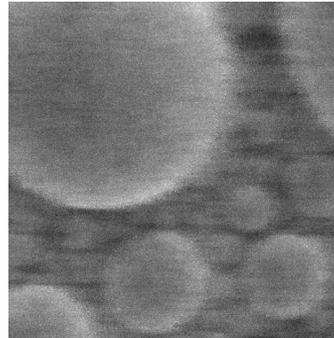
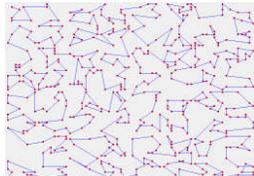
Visiting random positions



Pseudo-raster

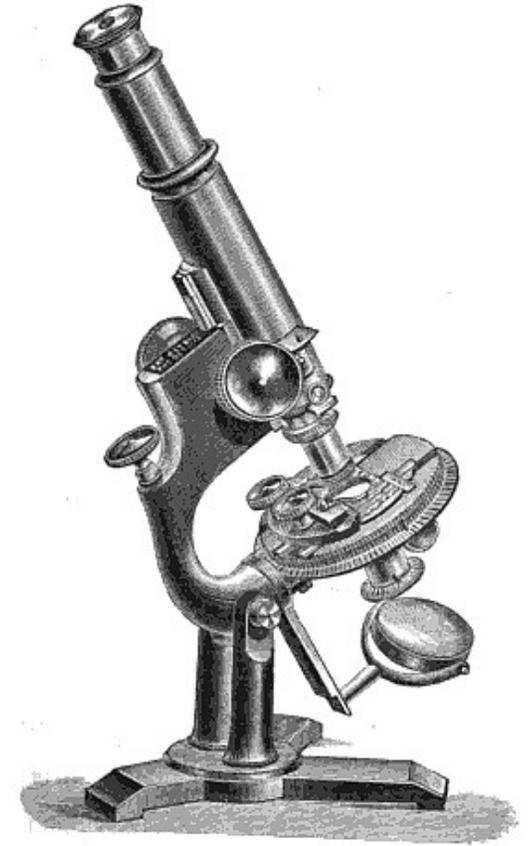


Minimum path scan



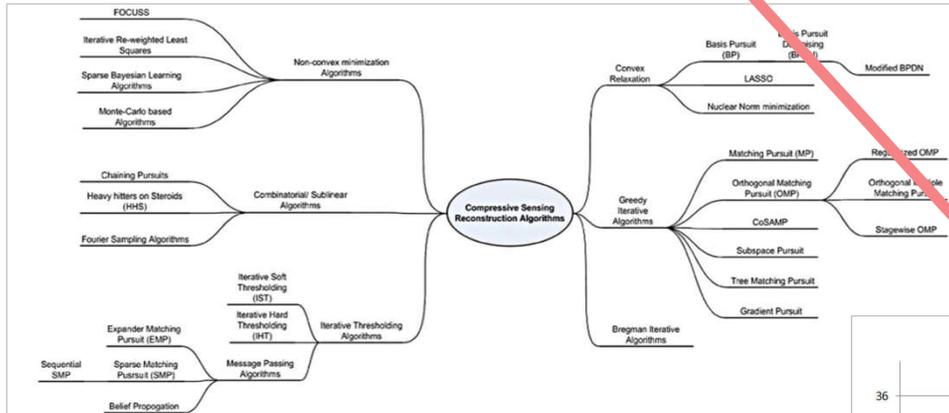
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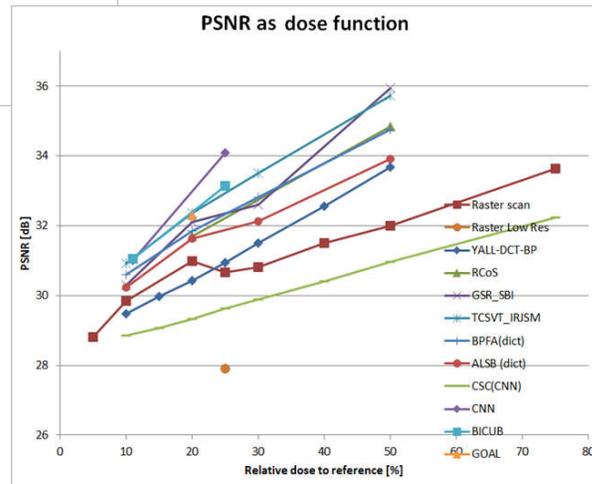
Reconstruction algorithms

Too many in the air ...

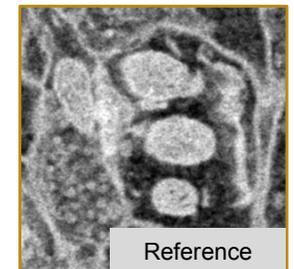
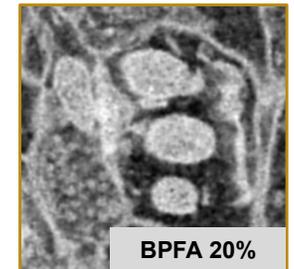
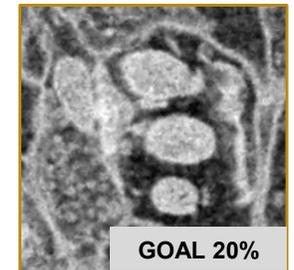


Signal characteristic(s)	Sparsifying transform ψ
Locally periodic	Discrete cosine (DCT)
Periodic	Fourier (DFT)
Piecewise smooth	Wavelets
Piecewise constant	Finite differences (TV)
Specific	Custom dictionaries

Tested

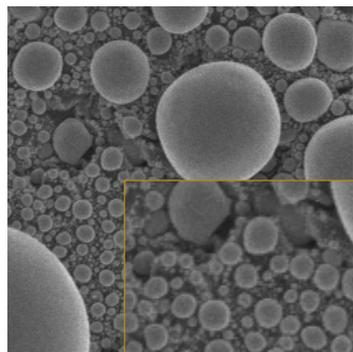


Selected

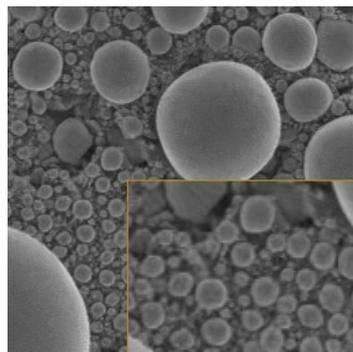


GOAL – Geometric Analysis operator Learning
BPFA – Beta Process Factor Analysis

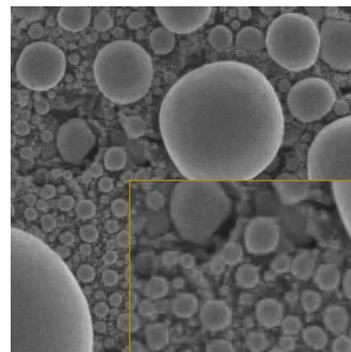
Reconstruction algorithm comparison at 50%



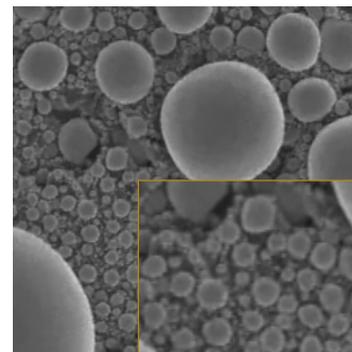
Reference
DT=1us



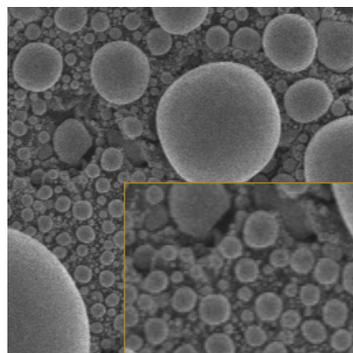
GSR_SBI



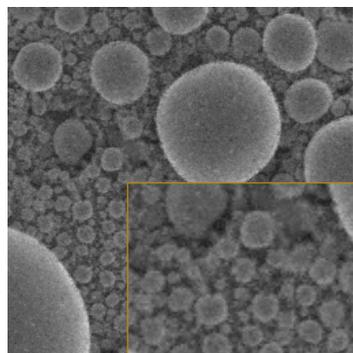
TCSVT_IRJSM



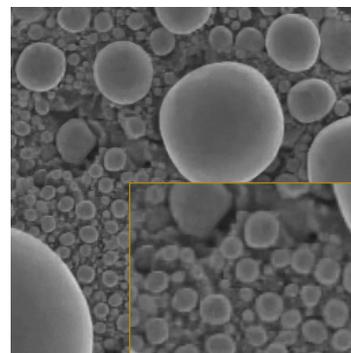
BPFA



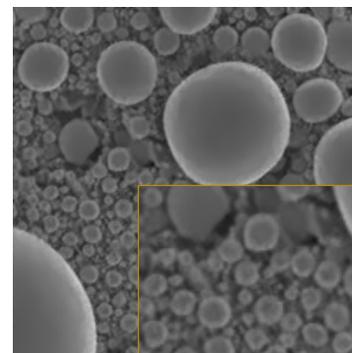
Raster 500ns



BP + DCT

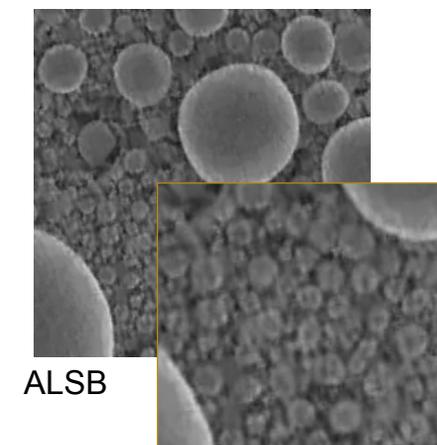
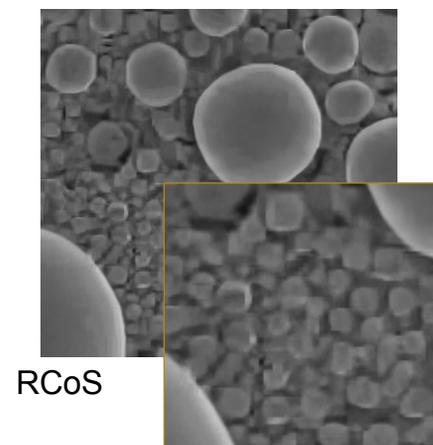
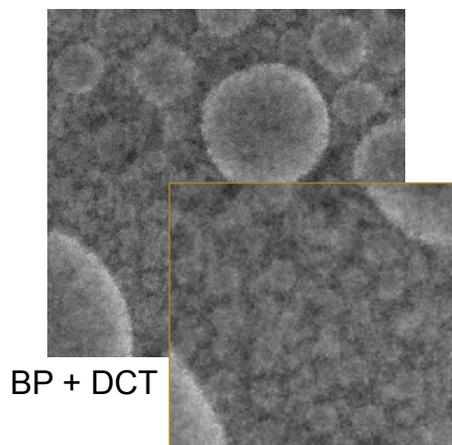
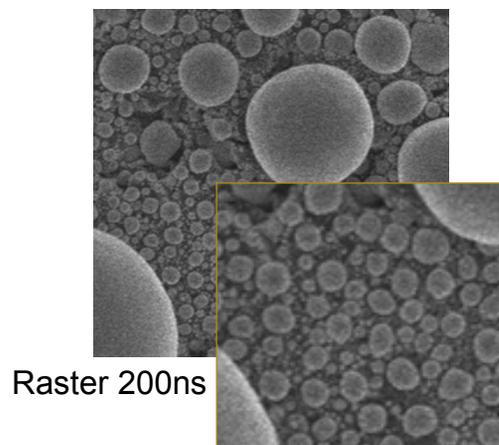
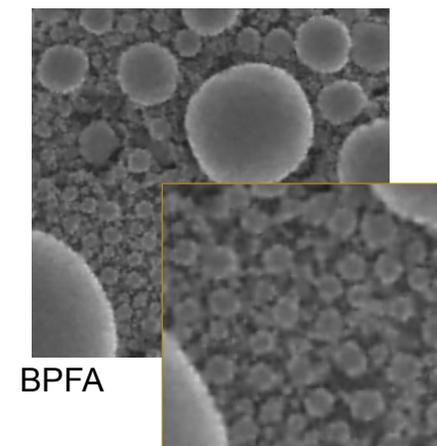
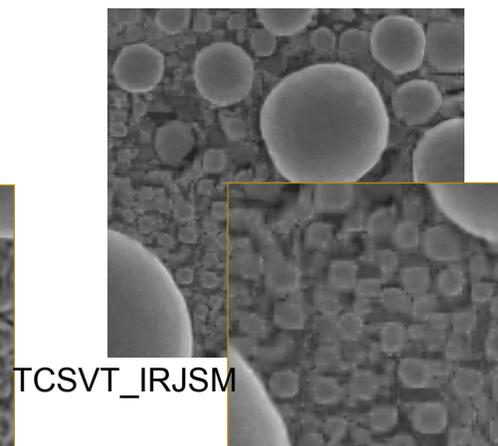
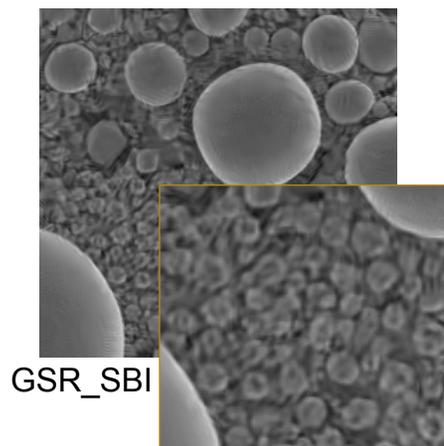
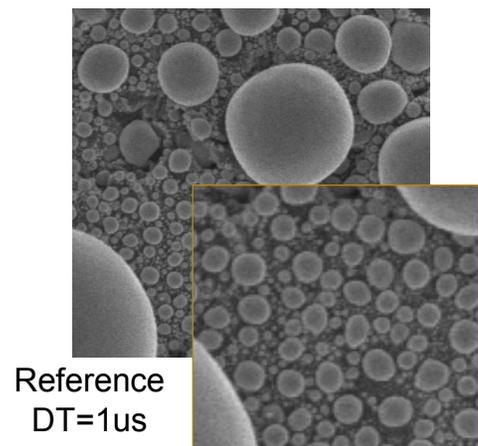


RCoS



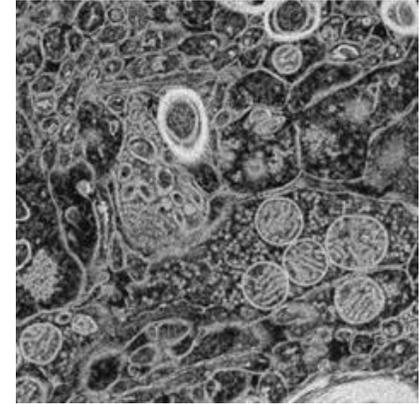
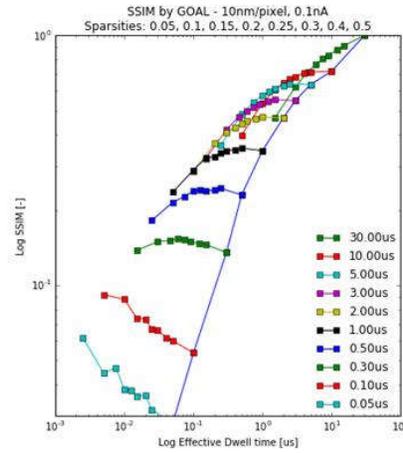
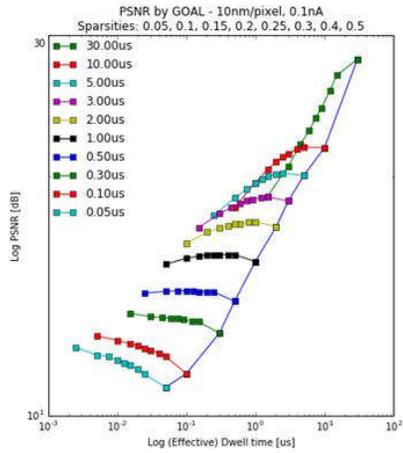
ALSB

Reconstruction algorithm comparison at 20%

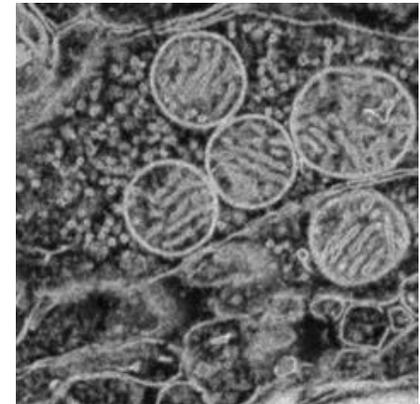
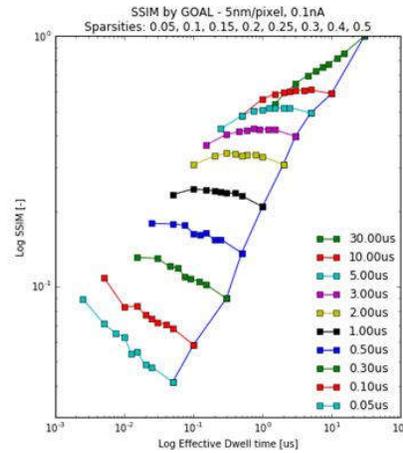
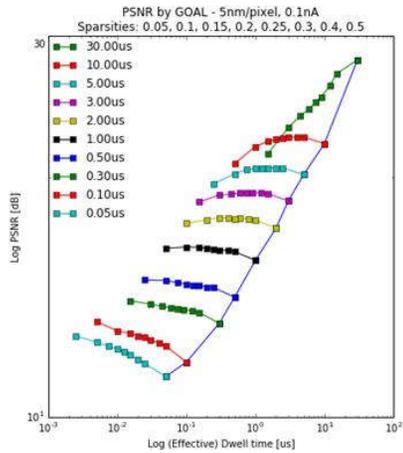


Dose-sparsity image quality analysis

10nm/pixel

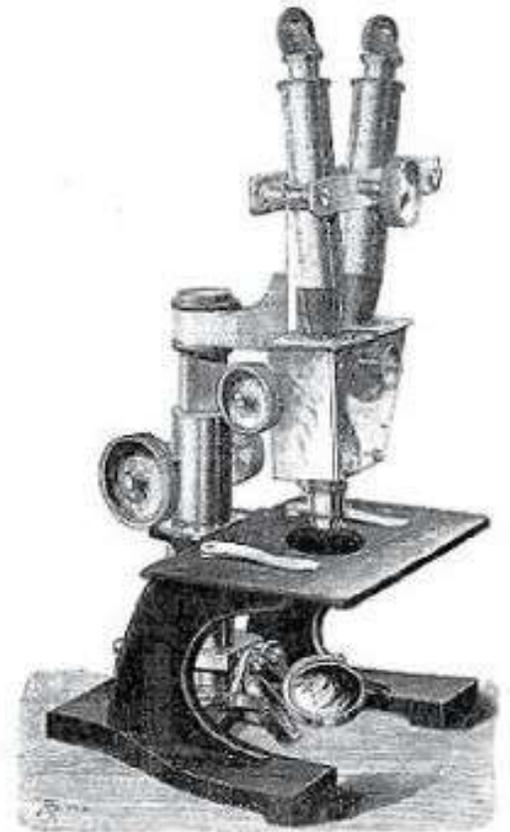


5nm/pixel

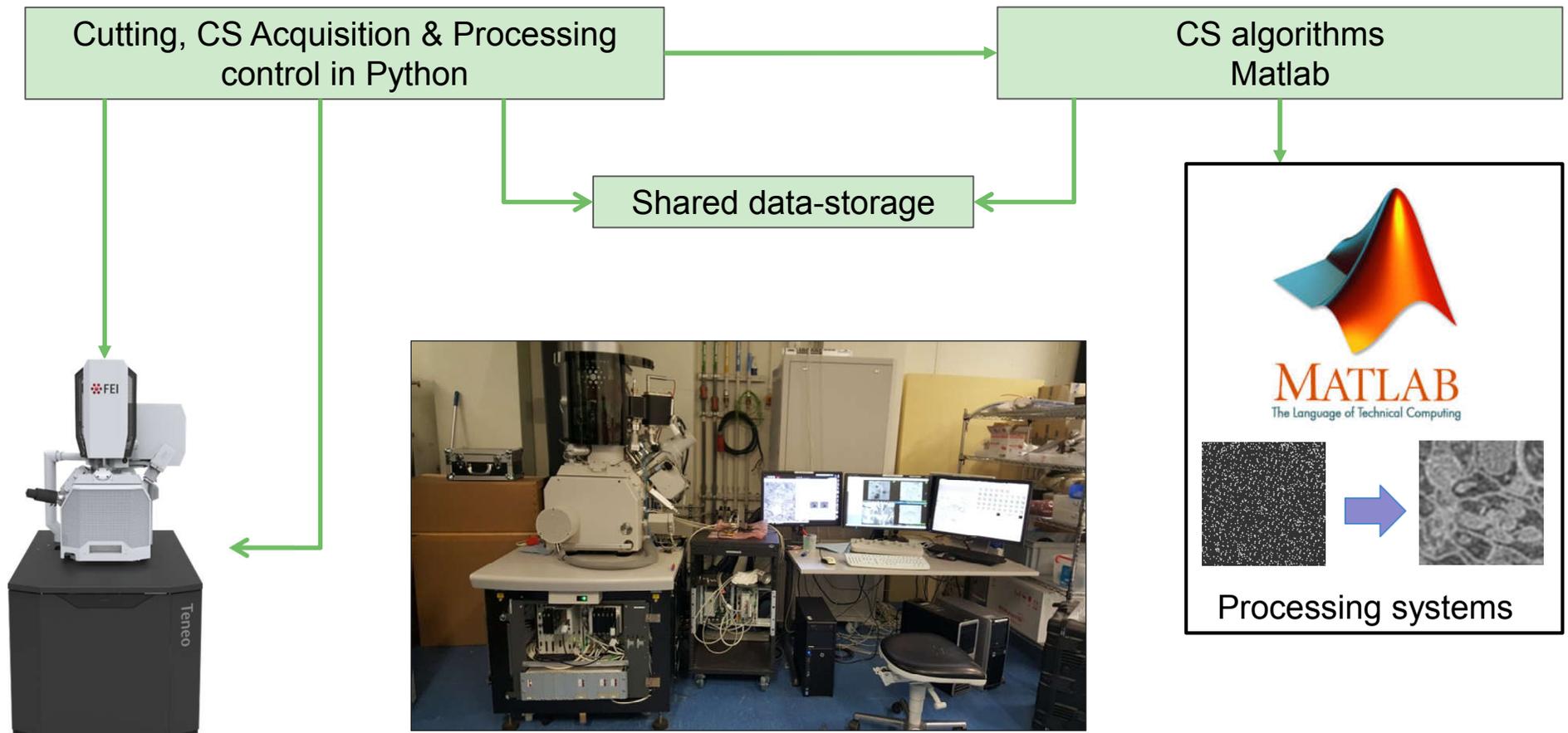


Overview – How we use Matlab for Big Data Acquisition

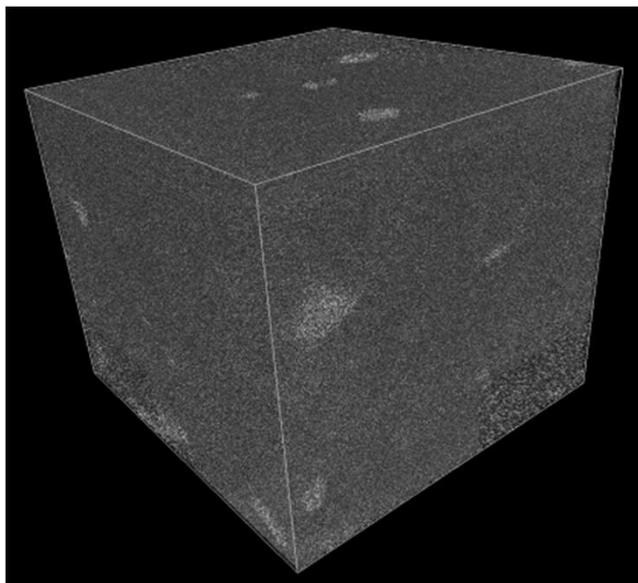
- **Life Science, Electron Microscopy**
- **Matlab Integration: Image Acquisition Stability**
- **Compressive Sensing Basics**
- **Image Acquisition with CS**
- **Matlab: Reconstruction Algorithms**
- **Matlab Integration: Integration to Acquire Big Data**



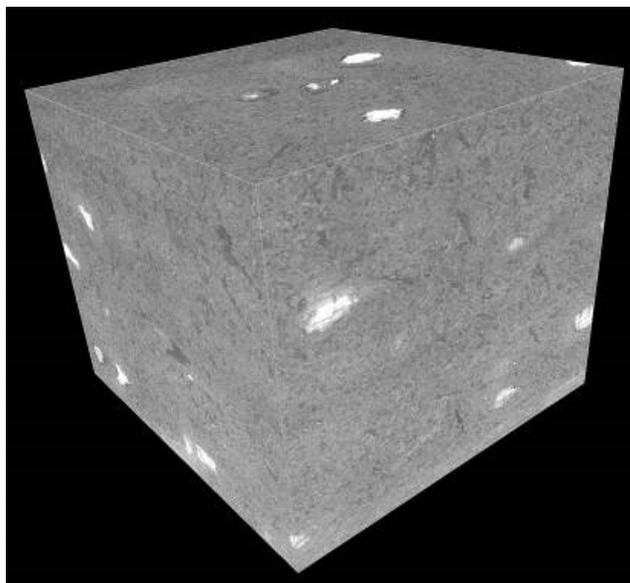
How Matlab helps to evaluate Compressive Sensing?



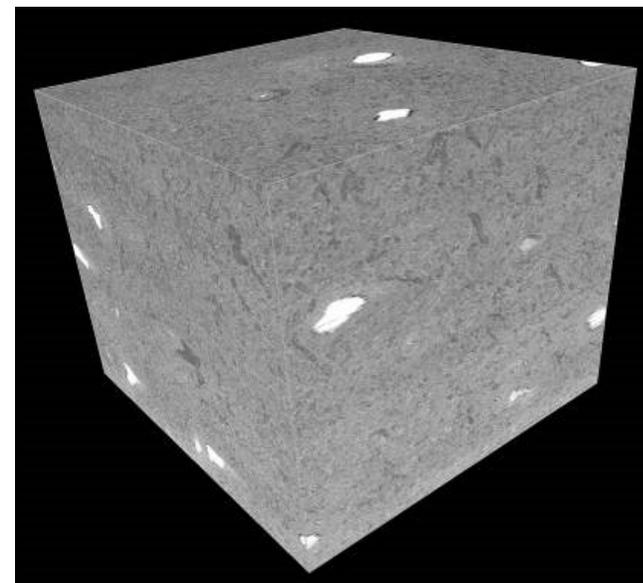
Large Dataset Acquisition - Comparison



Sparse Volume



Reconstruction



Full Grid Volume

To take away

- **Matlab is a nice computational tool**
- **It is even nicer when integrated in full workflow**
- **It is very good to evaluate algorithms**
(image sharpness, compressive sensing, ...)

